

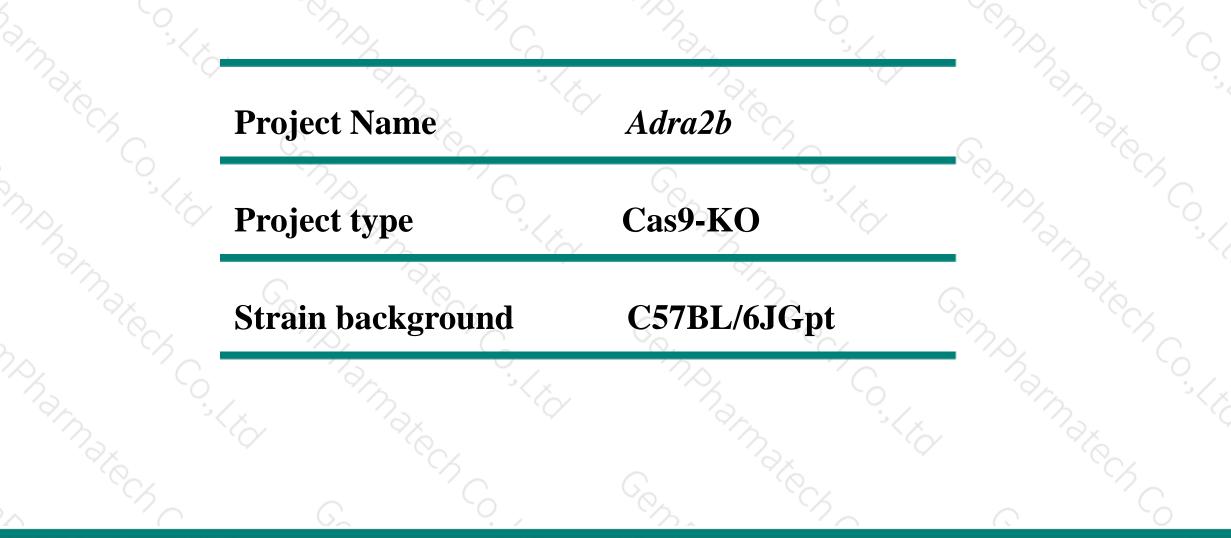
Adra2b Cas9-KO Strategy

Designer: Xiaojing Li Design Date: 2019-9-19 Reviewer: JiaYu

Project Overview



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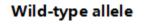


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This model will use CRISPR/Cas9 technology to edit the Adra2b gene. The schematic diagram is as follows:



KO allele



Uncoding region

AŢG

Coding region

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- The Adra2b gene has 2 transcripts. According to the structure of Adra2b gene, exon1 of Adra2b-201 (ENSMUST00000071902.4) transcript is recommended as the knockout region. The region contains all of the coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify Adra2b gene. The brief process is as follows: sgRNA was transcribed in vitro.Cas9 and sgRNA were microinjected into the fertilized eggs of C57BL/6JGpt mice.Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.





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- Homozygous null mice exhibit poor survival and breeding, lack the vasoconstrictor response to alpha2-adrenergic receptor agonists, and display background strain dependent postnatal respiratory failure. Heterozygotes show an attenuated hypertensive response to subtotal nephrectomy and salt loading.
- The Adra2b gene is located on the Chr2. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risk of the gene knockout on gene transcription, RNA splicing and protein translation cannot be predicted at the existing technology level.

Gene information (NCBI)

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Adra2b adrenergic receptor, alpha 2b [Mus musculus (house mouse)]

Gene ID: 11552, updated on 12-Aug-2019

Summary

Official SymbolAdra2b provided by MGIOfficial Full Nameadrenergic receptor, alpha 2b provided by MGIPrimary sourceMGI:MGI:87935See relatedEnsembl:ENSMUSG00000058620Gene typeprotein codingRefSeq statusVALIDATEDOrganismMus musculusLineageEukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia;
Myomorpha; Muroidea; Murinae; Mus; MusAlso known as[a]2B; a2b-AR; Adra-2b; alpha2B; alpha2-C2
human all

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Transcript information (Ensembl)



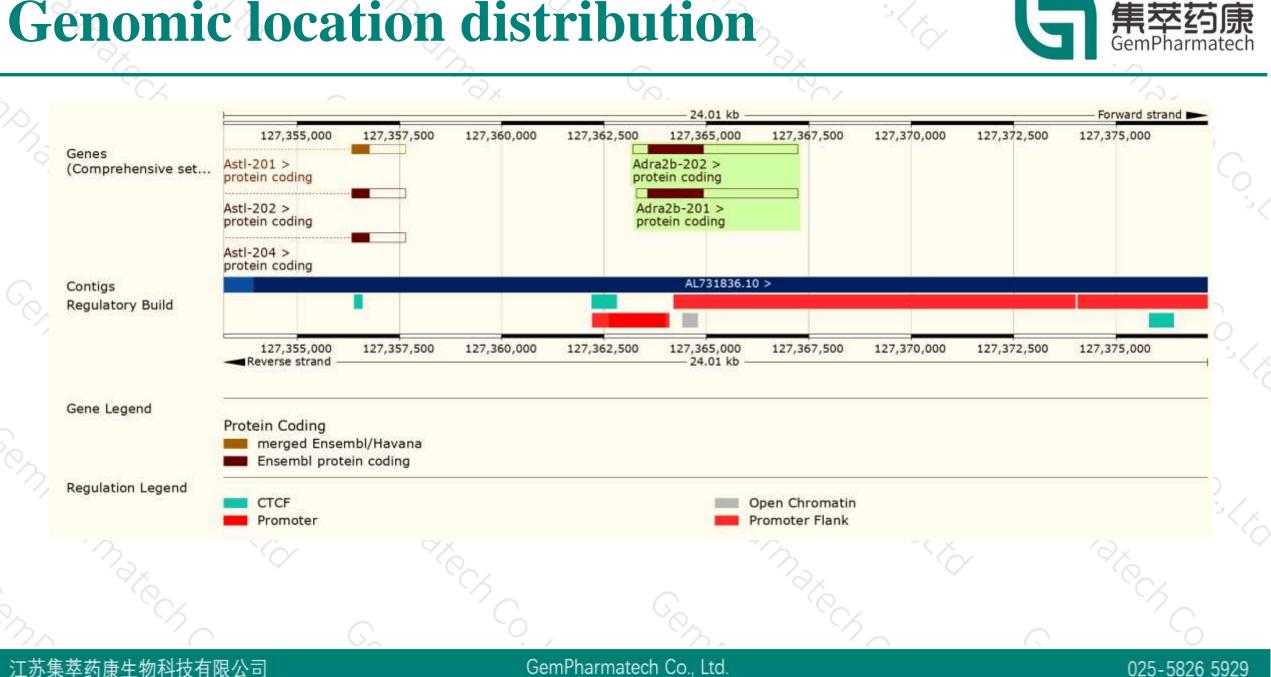
The gene has 2 transcripts, and the transcript is shown below:

Name 🔺	Transcript ID 💧	bp 🖕	Protein 🖕	Biotype 🍦	CCDS	UniProt 🖕	Flags	\$
Adra2b-201	ENSMUST0000071902.4	3936	<u>453aa</u>	Protein coding	<u>CCDS16701</u> @	<u>F8VQ23</u> &	TSL:NA GENCODE basic	APPRIS P2
Adra2b-202	ENSMUST00000104934.1	4014	<u>448aa</u>	Protein coding	-	<u>Q925K6</u> മ	TSL:NA GENCODE basic	APPRIS ALT2

The strategy is based on the design of Adra2b -201 transcript, The transcription is shown below

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	Adra2b-201 > protein coding	10					
	Adra2b-201 >			3.94 kb —		Forward strand F	
		*. /	()		(-)		

Genomic location distribution



Protein domain

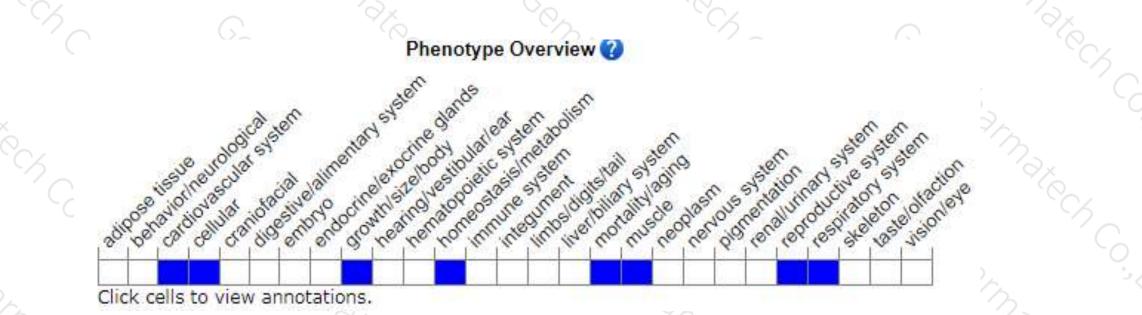


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	Prints		pled receptor, rho		_									5
	Francis	G protein-coupled	receptor, rhodop	sin-like									-	
			Adrenocepto	r family		0,000		Alpha 2B a	adrenoceptor			0	-	
	Pfam	G protein-c	oupled receptor, r											
	PROSITE profiles	GPCR, rhod	opsin-like, 7TM											
	PROSITE patterns			G protein-coupled	receptor, rhod	opsin-like								
	PANTHER	PTHR24248											- Š	
		PTHR24248:SF130												
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Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue. Data quoted from MGI database(http://www.informatics.jax.org/).

Homozygous null mice exhibit poor survival and breeding, lack the vasoconstrictor response to alpha2-adrenergic receptor agonists, and display background strain dependent postnatal respiratory failure. Heterozygotes show an attenuated hypertensive response to subtotal nephrectomy and salt loading.

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If you have any questions, you are welcome to inquire. Tel: 025-5864 1534



